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## Review

# Effects of high-pressure processing (HPP) on the microbiological, physico-chemical and sensory properties of fresh cheeses: A review

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High pressure processing (HPP) is an increasingly popular food processing method that offers great potential within the food industry. The drive to use HPP is to provide minimally processed foods which are safe and have extended shelf-life that rival traditional methods of food processing. HPP is currently being applied to a wide variety of food products, although to date the dairy industry has received little attention. The present paper reviews the effects of HPP on fresh rennet- and acid-coagulated cheeses. In addition to modifying physicochemical and sensory characteristics, HPP is reported to inactivate certain micro-organisms typically found in cheeses. Pathogenic microorganisms such as *Listeria monocytogenes* and *Escherichia coli* which contaminate, spoil and limit the shelf-life of cheese can be controlled by HPP. HPP can also cause changes in milk rennet coagulation properties, produce a more continuous or homogeneous protein matrix in cheese, improve cheese structure, texture and yield, as well as reduce moisture content variations within fresh cheese blocks. Providing HPP can be operated economically, the use of pressure may be an attractive new method for the processing of cheese.

**Key words:** High pressure processing, fresh cheese, dairy, spoilage.

## INTRODUCTION

Food processors aim to satisfy consumer demands for foods that are fresh, attractive and present no risk to health (Trujillo et al., 2000). Minimally processed foods are increasingly attractive to the consumer. Non-thermal food preservation methods which are receiving much interest include high-intensity light pulses, ionizing radiation and high hydrostatic pressures (Valero et al., 2007). Sensory, biochemical, microbial and physical parameters are used to define a product's freshness (Karoui et al., 2006), while psychological attitude, cultural, religious factors as well as consumption rate and their regional preferences as well as methods of preservation can determine a product's acceptability and quality (Fox et al., 2004).

High pressure processing (HPP) is an isostatic non-thermal pressure method that is applied batch-wise at typical pressures ranging from 200 to 600 MPa. From the pioneering days of examining the effects of pressure on food, recent developments are the consequence of advances in equipment fabrication. Today, however, HPP products are available in Europe, Japan and USA. Examples include fruit juices, meats, seafood, avocados and salsa amongst others. Unlike thermal processing of foods, HPP provides a uniform and instantaneous effect without the need for heating and may be carried out at ambient temperatures (Patterson, 2005). HPP is capable of modifying the structure of food macro-components which contain proteins through the disruption of non-covalent interactions causing minimal changes to food characteristics and quality (Cheftel, 1991; Huppertz et al., 2002). Temperature may also cause denaturation effects although the mechanisms are somewhat different. The duration of the exposure to both pressure and temperature

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is also known to have a marked influence (Mertens and Knorr, 1991). HPP can be used alone or in combination with other processing methods to enhance the overall or desired effects (Thakur and Nelson, 1998; Rastogi et al., 2007).

Trujillo et al. (2002) has previously summarized the potentials of pressure-induced modifications of dairy products. There is, however, no comprehensive review which documents the effects of HPP on the microbiological, physico-chemical and sensory properties of fresh cheeses. In this review, we present the avenues through which the quality of fresh cheeses can be improved by HPP including rennet coagulation, physico-chemical, sensory and textural properties, inactivation of microorganisms, protein effects, moisture content, cheese yield and salting.

## CHEESE: PROBLEMS AND SPOILAGE

Cheese is a nutritious and versatile dairy food manufactured from buffalo, cattle, goats and sheep milk. It is estimated that there are in excess of five hundred distinct cheese varieties are currently produced worldwide (Fox et al., 2004). Cheese makers strive to maintain consistent and uniform raw material quality in cheese production (Karoui et al., 2006). Factors such as consistency variations, production and quality affect the flavour, texture and appearance of fresh cheese. Major steps in fresh cheese include assembling of the raw milk, formation of the cheese milk, formation of the coagulum, formation of the curds, before final fresh cheese. The key points include:

1. Thermization (a less-severe heat treatment than pasteurisation which reduces microbial population during cheese-making), standardisation and pasteurization;
2. Starter and secondary cultures present in the cheese milk;
3. Cutting, cooking (which accounts for acidification, composition and syneresis of curd that would be formed as well as the retention of the coagulant), agitation and draining of coagulum and
4. Acidification, dehydration, salting, moulding and pressing of curd, after which a 'fresh' or 'soft' cheese emerges (Fox et al., 2004).

Fresh cheese can be faced with problems during manufacture as well as storage. Cheese spoilage is complex with losses even with today's preservation methods. Contamination of fresh cheese can occur during processing and shelf-life, followed by serious deteriorations. Pathogenic microorganisms such as *Listeria monocytogenes* and *Escherichia coli* are known to spoil fresh cheeses (Trujillo et al., 2000). Sporadic reports of listeriosis (Bell and Kyriakides, 2005) caused by milk-borne bacteria *L. monocytogenes* trouble the cheese industry. *Listeria* out-

breaks on soft cheeses have resulted in severe economic impact and mortality (Arqués et al., 2005; Wagner and McLauchlin, 2008), though *Listeria* contamination in cheese processing plants is uncommon (Pak et al., 2002). *L. monocytogenes* in ready-to-eat foods [RTE] must be controlled and this can be achieved since the microorganism can thrive at above pH 6 and poorly or not at all on products near or below pH 5 (Glass and Doyle, 1989). The occurrence of spoilage yeasts can limit the refrigerated shelf-life of certain fresh cheeses. Excess acidification can also adversely affect a cheese's quality (Daryaei et al., 2008).

## HIGH-PRESSURE PROCESSING OF CHEESE

Areas of interest of HPP on cheese manufacture include rennet coagulation of milk, its impact on physiochemical and sensory properties of cheese and the inactivation of contaminating microorganisms. Trujillo et al. (2000) noted that pressure, duration of exposure and temperature which when used in combination provide the determinants for producing optimal effects on processed fresh cheese.

### Rennet coagulation properties

Rennet coagulation is a two-stage process. The primary stage is an enzymatic hydrolysis of Phe<sub>105</sub>-Met<sub>106</sub> bond of  $\kappa$ -casein ( $\kappa$ -CN) by chymosin which destabilizes the casein micelles to yield a *para*- $\kappa$ -casein and casein macropeptides. This is followed by a secondary stage which involves conglomeration of modified micelles by enzymes. Coagulation process and cheese making properties of milk are indirectly influenced by HP treatment of milk through a number of effects on milk proteins such as reduction in the size of casein micelles and denaturation of  $\beta$ -Lactoglobulin ( $\beta$ -Lg), probably followed by interaction with micellar  $\kappa$ -casein (O'Reilly et al., 2001).

Rennet coagulation time (RCT) of cow's milk is influenced by HP treatment. The nature and magnitude of the effect is said to depend on the pressure applied (O'Reilly et al., 2001). There are a number of factors which influence rennet coagulation properties of milk. The reversible nature of HP-induced changes enables casein dissociation without being accompanied with any increased Ca<sup>2+</sup> ion activity (López-Fandiño, 2006a). This phenomenon has been observed within range of 100 - 400 MPa on fresh cheese in which rennet coagulation properties have been affected because micellar  $\kappa$ -casein dissociated due to the activity of  $\beta$ -Lactoglobulin ( $\beta$ -Lg) and casein micelles. The reversible casein dissociation was found to increase the rate and strength of gel formation in HPP milk (100 – 400MPa) without increasing Ca<sup>2+</sup> activity – the main mechanism underscoring reduction in RCT (Johnston et al., 2002; Zobrist et al., 2005).

Using a processing time less than 5 min and temperature of 21°C, pressures from 100 to 600 MPa applied on raw milk brought about rennet coagulation followed by a decrease in RCT. Denaturation of whey proteins was suggested to be the reason of this occurrence (Huppertz et al., 2005; López-Fandiño, 2006b). Heated milk treated with pressures approaching 600 MPa (0 - 30 min) showed equal to lower RCT than non-treated milk. Gel strength of the rennet-induced coagulum from heated milk treated at 250-600 MPa for 30 min or 400 or 600 MPa for 0 min was found to be higher relative to that of unheated unpressurised milk. Also, HP with heat treatment was viewed as commercially attractive since desirable effects on rennet coagulation and cheese-making properties could be obtained at short time intervals (Huppertz et al., 2005). Trujillo et al. (1999b) compared the coagulation properties of pasteurised (72°C for 15 s) and HP treated (500 MPa for 15 min at 20°C) milk. They obtained a statistically significant difference ( $p < 0.05$ ) between the RCT of HP-treated milk (which was higher) when compared to pasteurised milk. Lopez and Olano (1998) examined the influence of HP on the rennet coagulation properties of ewe and goat's milk and found that treatment at 400 MPa did not extend noticeably the coagulation time. Particularly, the consistency of the curd was improved in the case of the goat's milk.

Separate effects of HP treatment on the two stages of rennet coagulation of milk have been examined by some workers. López-Fandiño et al. (1997) and Trujillo et al. (1999b), were both with similar opinion about the negative influence of HP treatment on the aggregation stage at pressures greater than 300 MPa. They found that the negative influence may be as a result of the association of denatured  $\beta$ -Lg with casein micelles which interfere with the aggregation of the micelles. A possible alternative mechanism was suggested by Needs et al. (2000) such that large oligomers of denatured  $\beta$ -Lg formed in the milk's serum phase, interfered physically with the aggregation of casein micelles. This phenomenon provided indirectly somewhat supporting evidence for the above hypothesis made by López-Fandiño et al. (1997) and Trujillo et al. (1999b).

### Physicochemical, sensory and textural properties

HPP at 500 MPa for 5 to 30 min at 10 or 25°C was applied on a fresh cheese made from goats' milk. HP treatment did not change the composition of fresh cheese especially the total solid (TS), ash, fat and soluble nitrogen (SN) contents. However, non-protein nitrogen (NPN) of HPP cheeses remained lower than the non-treated fresh cheese. Clearly, the changes in water retention capacity were minor, but did not affect the nutrient content of the cheese. However, total nitrogen (TN) increased in the whey of HPP cheese followed by increased

whey loss compared with control cheese (Capellas et al., 2001). Physicochemical findings of Capellas and co-workers focused on a different aspect compared with Trujillo and co-workers. In the studies of Trujillo et al. (1999 a,b), when HP treated milk cheese was compared with pasteurised milk cheese (control), higher moisture, salt and total free amino acid content was obtained over the non-processed milk cheese.

With HPP (200 MPa, 20°C), functional properties of immature cheese showed significant changes in moisture redistribution within the cheese microstructure. This was further exemplified by the serum lightness and yellowness values (Johnston and Darcy, 2000). No significant effect on composition, pH and proteolysis of reduced-fat cheese occurred at HPP of 400 MPa for 5 min at 21°C, instead  $L^*$ -value (whiteness) decreased with yellowness while greenness increased as tempering at 21°C for 16 h occurred on 'day one' of the cheese (Sheehan et al., 2005). Using HPP of 500 MPa for 5, 15 and 30 min at 10°C (PT1) or 25°C (PT2) (PT = pressure-treated), colour parameters hardly changed in the inner part of a fresh cheese although surface modifications were more pronounced. Here, total colour difference ( $\Delta E^*$ ) values of PT1 cheeses were greater than PT2 mainly due to  $L^*$ -value changes which occurred in HPP samples treated at 10°C. Also,  $b^*$  value was the index that changed in all treatments because it increased in PT- cheeses, that is, approached yellowness, as treatment time increased. The study suggested that increase in lightness and yellowness of the cheese surface was likely to relate with changes in cheese microstructure (Capellas et al., 2001).

Rheology or cooking properties of the cheese after HPP (400 MPa for 5 min at 21°C) appeared not to be significantly affected during storage (Sheehan et al., 2005). HPP milk cheese (483 MPa at 10°C) using low (2.75 rad/s) to high (173 rad/s) angular frequencies and HPP milk cheese (483 MPa HP at 10°C) using creep tests of minimal instantaneous compliance ( $J_0$ ), both revealed a more solid-like property of cheese compared to their control (non-treated pasteurized-milk) cheeses. While deformability of pasteurized milk cheeses (control) increased, a decline in the solid-like property of control followed when compared with HPP-milk cheeses as shown by the  $J_0$  value (San Martín-González et al., 2007). Confocal scanning light microscopy (CSLM) enables microscopic observation of changes that occur in fresh cheese treated with HPP. When HP-treated cheese was compared with the non-pressurised cheeses, a more continuous/homogeneous protein matrix was obtained. The fat phase was observed to be equally affected, most of which occurred at pressures up to 450 MPa and exposure of 15 min upwards. Also, the circularity of the fat globules was lost and large pools of fat were obtained (O'Reilly et al., 2001). However, no difference was observed in CSLM when the non-processed fresh cheese microstructure was compared with fresh cheese treated with HPP of 500 MPa for 5, 15 and 30 min at 10 or 25°C

(Capellas et al., 2001). This may have occurred probably because the fresh cheese underwent slight pressing before pressurisation. High pressure homogenized the protein structure and altered the fat globule distribution of cheeses not pressed during cheese making (Capellas et al., 1997).

HPP milk cheese (500 MPa, 20°C) showed firmer, more elastic but less fracturable texture compared with pasteurized milk (72°C, 0.25 min) with cheese-matrix structure similar to the raw milk cheeses (Buffa et al., 2003). Cohesivity of the raw milk cheese increased in HPP over the pasteurized milk (Trujillo et al., 2002). Torres-Mora et al. (1996) pioneered the phenomena where the HP treatments (275 MPa for 100 s) applied to fresh curd immediately after manufacture obtained microstructural and textural changes within the blocks of HP treated cheese and that the texture attribute of processed fresh resembled aged Cheddar cheese. This finding was later confirmed by Yang (1999). HPP (full-fat) cheese (400 MPa; 10 min; room temperature) revealed a significantly high strain and stress fracture values and this was followed by low flowability a day after manufacture compared with the non-processed cheese (Rynne et al., 2008).

Hard surface which was evident in a HPP milk cheese (500 MPa in the range of  $30 \pm 5$  min at 25°C) was subjected to sensory analysis and the result was that texture attributes did not affect panelists' preference for processed cheese over the non-processed cheese (Trujillo et al., 2000). In contrast, HP treatment up to 500 MPa applied on cheese was reported to improve sensory characteristics of HP treated compared to untreated cheeses (Kolakowski et al., 1998). Appearance, smell or taste of the cheeses remained unaffected at HPP (200 - 500 MPa for 15 min) because changes between HPP cheese and control were undetected by panelists (Szczański et al., 1997). This tends to agree with the work of Daryaei and colleagues because no notable adverse effects on the sensory and textural quality were evident in a fresh (lactic curd) cheese when processed at HP treatment from 200 to 600 MPa (Daryaei et al., 2006).

### Inactivation of microorganisms

In recent times, the inactivation of microorganisms of fresh cheeses by HP treatment has received considerable attention. In these works, target microorganisms were typically inoculated in the cheese or milk, after which HP treatment was applied. Microorganisms which have been of primary focus for fresh cheeses include Gram positive microorganisms such as *L. monocytogenes*, *Staphylococcus* spp., *Bacillus* spp. and Gram negative microorganisms such as *E. coli*. Yuste et al. (2001) noted that the barometric sensitivity of microorganisms can vary to a high degree. Possibly, this accounts for the reason why the mechanism of inactivation

of microorganisms by HP treatment is not fully understood. Cell membranes of microorganisms become damaged, distorted and permeable as a response to the challenges of HP treatment and that brings about changes in morphology, cell membrane and wall, biochemical reactions and genetic mechanisms. Primarily, HP treatment targets cytoplasmic membrane of microorganisms and starts the crystallization of membrane phospholipids which lead to microbial inactivation. This HP-induced changes on cell membrane brings about sublethal injury to some microorganisms and as such, the latter become weakened and cannot grow on selective media, hence, need time to recover (Yuste et al., 2001).

Practical applications related to the inactivation of microorganisms are presented in Table 1. HPP (450 MPa/10 min or 500 MPa/5 min) on raw goat milk cheese destroyed the population of *L. monocytogenes* without significantly affecting sensory characteristics of cheese (Gallot-Lavallée, 1998). A 6 log<sub>10</sub> unit reduction of inoculated *L. monocytogenes* was reported in sliced cheese by HPP (500 MPa; 15 min) which accompanied by a significant decrease of cheese microbiota (Szczański et al., 1997). HP treatment with pressures up to 500 MPa for 10 min at 20°C demonstrated significant reduction of level of *L. monocytogenes* in the raw milk. This gave way for the production of a safer but non-thermally processed camembert-like soft cheese (Linton et al., 2008). A near 100-fold reduction in starter bacteria numbers followed by delayed growth of non-starter lactic acid bacteria (NSLAB) were attained in a 1 day-old full-fat cheese under HPP (400 MPa for 10 min at room temperature), one-day post-manufacture. Here, the depletion of the primary energy source (lactose), autolysis, as well as the inhibition by salt, were considered as factors which may have likely contributed to the reduction in bacterial numbers (Rynne et al., 2008). Inactivation of starter bacteria and spoilage yeasts was found in a vacuum packaged fresh cheese by HPP (200 to 600 MPa; 5 min;  $\leq 22^\circ\text{C}$ ). Shelf-life was increased to about 8 wk when HPP fresh cheese was stored at 4°C. HPP reduced the viable counts of *Lactococcus* in both aerobic and anaerobic incubation conditions, although yeast growth emerged at 200 MPa but was controlled at above 300 MPa (Daryaei et al., 2008).

A 7 log<sub>10</sub> unit reduction of *Escherichia coli* populations previously inoculated on vacuum packaged fresh cheese was evidenced at HPP of 400 to 500 MPa (2 - 25°C for 5 - 15 min). In cases where *E. coli* cells survived the HPP, the population remained at non-detectable levels after a week at 4°C. They neither developed nor survived the HPP treatment even after 2 month storage at 4°C (Capellas et al., 1996). Cocci (*Staphylococcus carnosus*) and spores (*Bacillus subtilis*) are microorganisms associated with fresh cheese and are pressure resistant. In HPP fresh cheese (400 - 500 MPa; 10 or 25°C; 30 min), no decrease in population counts of inoculated *S. carnosus* was observed even with the addition of mild

**Table 1.** Practical applications for inactivation of microorganisms in fresh cheeses by high pressure (HP) treatment.

Inactivation applications	Cheese type	HP conditions	References
Reduction of inoculated <i>E. coli</i> by 7 log cfu g <sup>-1</sup> units; Least detection of surviving cells after a refrigerated week (4°C) with no microbial development even after 2 months of storage.	Mató (fresh) cheese.	400 to 500 MPa/2 - 25°C for 5 – 15 min at room temperature	Capellas et al. (1996).
Inactivation of <i>L. monocytogenes</i> yielded 5.6 log cfu g <sup>-1</sup> reductions without significant effect on cheese sensory characteristics.	Cheese from raw goat milk	450 MPa/10 min or 500 MPa/5 min	Gallot-Lavallée (1998).
Inactivation of both inoculated and indigenous bacteria obtained as well as vegetative cells such as <i>S. carnosus</i> , <i>B. subtilis</i> and aerobic mesophilic bacteria (AMB), obtained range of 2.7 to 7 log cfu g <sup>-1</sup> reductions. Multiple-cycle HPP improved inactivation rate of <i>S. carnosus</i> ; Combined treatment of HPP+nisin inactivated cheese indigenous microbiota.	Fresh cheese from goat milk.	400 - 500 MPa (10 or 25°C) for 30 min and mild heat; Multiple-cycle of 500 MPa (15 - 30 min) with nisin; 60 MPa (40°C) for 210 min.	Capellas et al. (2000).
Inactivation of starter and non-starter lactic-acid bacteria (NSLAB) resulted to least mean counts 1.5 log <sub>10</sub> cycles lower in HPP cheese. Significant reductions in both starter and NSLAB populations occurred and near 100-fold reduction in starter bacteria numbers followed by delayed growth of NSLAB.	Day old full-fat cheese.	400 MPa for 10 min at room temperature.	Rynne et al. (2008).
Inactivation of starter bacteria and spoilage yeasts such as commercial <i>Lactococcus</i> spp. inoculated in milk: 1)Reductions of 2 - 7 log <sub>10</sub> (aerobic) and 3 - 7 log <sub>10</sub> (anaerobic) units of <i>Lactococcus</i> viable counts in the cheeses; and 2)Yeast growth was not significantly prevented at 200 MPa but was controlled at higher pressure regimes (≥ 300 MPa).	Commercially manufactured fresh cheese from a pasteurized bovine milk.	200 to 600 MPa for 5 min at ≤22°C	Daryaei et al. (2008).

heat whereas HPP of 500 MPa (50°C for 5 min) resulted in a 7 log<sub>10</sub> cfu g<sup>-1</sup> reduction of *S. carnosus* (Capellas et al., 2000). Repeated-cycle HPP (500 MPa; 15 - 30 min) was found to improve the inactivation rate of *S. carnosus*. However, the most effective treatment was a combination of HPP (500 MPa) with nisin and this brought about the inactivation of cheese indigenous microbiota (Capellas et al., 2000).

Cations present in milk e.g. Ca<sup>2+</sup> protect microorganisms against inactivation by HPP. To solve the problem, an improved inactivation rate was attained in indigenous as well as inoculated microorganisms of fresh cheese, especially the Gram-negative bacteria, by HPP combined with mild heat or nisin (Hauben et al., 1998). Microbial quality of HPP milk cheese (500 MPa for 15 min at 20°C) was similar to pasteurised milk (72°C for 15 s) cheeses (Buffa et al., 2001). HPP (500 MPa at 25°C during 5 or 30 min) increased the shelf-life of refrigerated

(4°C) vacuum-packaged fresh cheese to 2 or 3 months (Trujillo et al., 2002).

### Protein effects

Denaturation of protein by high pressure processing (HPP) is well known. Pressure and temperature applied followed by treatment time determine degree of protein denaturation (Huppertz et al., 2004). Denaturation activity which occurred in HPP (300 or 400 MPa respectively for 30 min at 20 - 25°C) milk resulted in the whey being incorporated into the curd (López-Fandiño et al., 1996). Bilbao-Sáinz and co-workers applied HP treatment (300 – 450 and 600 MPa) on milk with minimal thermal (about 40°C) effect and determined the protease enzyme-activity of the milk. Here, protease was found to be very resistant to high pressures, but pressure stability was greater in

raw milk compared with pasteurized milk, although the homogenization of milk appeared to demonstrate a protective effect on the enzyme (Bilbao-Sáinz et al., 2009).

Under HP treatment of 100-300 MPa, proteins can, but do not always, denature, dissolve or precipitate reversibly (Thakur and Nelson, 1998). HPP (100 MPa) increased protein denaturation temperature in a fresh cheese but declined at above 100 MPa (Rastogi et al., 2007). HPP milk cheeses showed rapid breakdown rate of protein constituents compared with pasteurized milk cheeses (Molina et al., 2000). This must have led to the rapid speed-up and subsequent emergence of the resultant flavour and texture. HPP cheese of 676 MPa at 10°C obtained a mean recovery value of 79.9% of protein content compared to recovery of 81.55% at 483 MPa at 30°C with no significant difference ( $p < 0.05$ ) between recovery obtained for the two treatments. Also, there was significant increase in protein retention compared with cheeses made from raw or pasteurized milk (San Martín-González et al., 2007).

Change can be induced in protein structure at high pressures above 200 MPa, while above 500 MPa can bring about non-reversible effects such as the unfolding of monomeric proteins, aggregation and formation of gel structures (Iametti et al., 1997). Hayakawa and colleagues monitored the HPP effects on the  $\alpha$ -helix content of  $\beta$ -Lg (Hayakawa et al., 1996). A complete breakdown of the  $\alpha$ -helix content of ovalbumin was obtained when pressures of 1000 MPa for 10 min were applied, but,  $\beta$ -Lg was reduced by approximately 90%. In these experimental conditions, the pressure-induced denaturation was reported to be more severe than the temperature-induced denaturation. Denaturation of proteins by HPP, however, does not resemble temperature-induced denaturation which is often irreversible because of the break-age of covalent bonds and/or aggregation of unfolded protein (Mozhaev et al., 1996). The exact unfolding denaturation mechanism of the HP-induced protein is not yet fully understood. High-pressure is said to induce modification of the structure of the protein molecules. At the molecular level, the protein structure which emerges is reported to look like a protein-folding intermediary and is characterised by the presence of a native-like degree of secondary structure but consists of a loss of tight packing within the hydrophobic core of the protein. This situation is said to bring about increased interaction with hydrophobic probe molecules such as anilino-1-naphthalene-8-sulphonic acid. However, pressures above 300 MPa can initiate a change in the tertiary structure such that hydrogen bonds become modified leading to a complete disruption of the hydrophobic bonds, which results in a change in the protein's reactivity (Chapleau et al., 2003).

Pressurization can induce modification in the protein network of cheeses. This can also be associated with changes that occur in protein matrices especially changes in objective and subjective textural properties. The

evidence of these were found in the microstructure of HPP goat's milk cheese (450 MPa at 10°C for 5 - 30 min) which exhibited under CSLM, a more continuous and homogeneous protein matrix than non-pressurized cheeses. Also, adhesiveness and tactile oiliness were both linked to the amount of oil on the cheese strip surfaces and this occurred probably because the fat phase was observed most affected by HPP (450 MPa,  $\geq 15$  min) (Capellas et al., 1997). When HP treatment (400 MPa for 10 min at room temperature) was applied on a 1 day old full-fat cheese, it was found that HP treatment showed little to no effect on primary proteolysis in cheese. This was consistent with the plasmin and chymosin activities measured and these two activities (that is, plasmin and chymosin) were unaffected by HP treatment (Rynne et al., 2008).

### Moisture content, cheese yield and salting

HPP (50 - 400 MPa with holding time ranging 20 - 100 s) was suggested to reduce moisture content variations within a block of cheese or among several blocks of cheese (Torres-Mora et al., 1996). Pasteurized or raw milk cheeses showed no differences in moisture content with HPP milk (500 MPa, 15 min, 20°C) cheeses (Trujillo et al., 2002). On the other hand, increased moisture content was reported in HPP milk cheese at three 1-min cycles of above 500 MPa over raw or pasteurized milk cheeses (Drake et al., 1997) although moisture retained in HPP cheese increased at 483 to 676 MPa at 30 - 40°C (San Martín-González et al., 2007). Higher moisture content, which was found to occur in HPP milk cheese, was consistent with the higher level of denatured whey protein retained in the cheese (Trujillo et al., 2000). However, HP-treated cheeses at 300 MPa for 10 min were found to have better water retention properties compared with non-treated cheeses (Juan et al., 2008). The findings obtained by Juan and co-workers seemed to go along with works of Messens et al. (2000), because the later workers showed that high moisture content occurred in Père Joseph goat's milk cheese probably as a result of HP-induced changes in the cheese protein network which resulted in an alteration of the cheese structure and thus enabled better retention of water in cheeses.

HPP on the milk before cheese making has been considered to directly influence cheese yield. Huppertz et al. (2004) showed that HP treatment induced changes in curd yield. Although curd yield was not influenced by treatment at pressures  $\leq 250$  MPa, however, pressures from 250 MPa and above obtained increased curd yield of about 25%. Alongside increased curd yield, increased curd moisture content and reduced protein content in the whey were found to have occurred under such pressures. It was suggested that increased curd yield probably occurred because of two possibilities: (a) incorporation of denatured whey proteins and (b) enhanced moisture

retention in the curd.

A shift in salt content of HPP milk cheese can occur and this is consistent with whey-brine interchange which takes place because of increased curd formation (Trujillo et al., 2000). Pressure brining at 300 MPa was reported to disrupt the *para*-casein micelle structure and thus, higher amount of nitrogen was found in the cheese serum of pressure-brined cheese compared with untreated cheese (Messens et al., 1998). Furthermore, when salt and water content analyses in cheese cylinders which were brined at high pressure (100 - 500 MPa) at ambient temperature was carried out, the total amount of salt taken up by the cylinders was not influenced by pressure brining. Instead, the water loss of the cheese cylinders was reduced at pressures from 200 to 300 MPa upwards (Messens et al., 1999).

## CONCLUSIONS

There have been considerable advances and commercial applications of high pressure treatment of foods over the past twenty years. HP treatment when applied to fresh cheese can influence its microbiological, physicochemical and sensory characteristics. There is a growing body of literature and knowledge concerning the effects of HPP on the microbiology and physico-chemical properties of fresh cheeses. However, there seems to be a paucity of knowledge connected to the sensory aspects of HPP cheese manufacture.

Understanding the science that brings about influence of HP treatment on the properties of fresh cheeses has been somewhat challenging. Investigations attempted by several workers have put forward research findings which seemed to target HP treated fresh cheese with optimized properties of flavour, taste and texture through improved coagulation as well as elimination of microbial deterioration at storage. However, overcoming the engineering challenges of HP treatment on cheese production is yet to be fully realized. Once unequivocal evidence is presented which demonstrates the economic advantages and benefits to be gained from HPP fresh cheese, HPP users and cheese manufacturers may turn their attention to this relatively new food processing technique.

While these investigations go on, cheese makers need to increase and strengthen the distribution of their local cheese products, pursue possibilities to extend shelf-life of cheese products and to explore further the yet-to-be commercialized aspects of the cheese industry. Possibly, at a point, they would appreciate and realize the significance of HPP.

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